

CLAIMS

We claim :

1. An optical labeling molecule comprising:
 - a. a zwitterionic dye moiety;
 - b. a titratable group moiety; and
 - c. a functional linker moiety.
2. The labeling molecule of Claim 1 further comprising a cleavable moiety.
3. The labeling molecule of Claim 1 or 2 further comprising a second label.
- 10 4. The labeling molecule of Claim 3, wherein the second label is a light stable isotope label.
5. The labeling molecule of Claim 3, wherein the second label is a heavy stable isotope label.
6. The labeling molecule of Claim 1, wherein charges on the zwitterionic dye moiety are stable between pH 3-12.
7. The labeling molecule of Claim 1, wherein the linker is an amine-reactive linker.
- 15 8. The labeling molecule of Claim 1, wherein the linker is a thiol-reactive linker.
9. The labeling molecule of Claim 1, wherein the linker is selected from the group consisting of amino group reactive imidoesters, N-hydroxysuccinimidyl esters or sulphydryl-reactive maleimides or iodoacetamides.
10. The labeling molecule of Claim 1, wherein the zwitterionic dye moiety comprises a BODIPY dye
20 with at least one zwitterionic component.
11. The labeling molecule of Claim 1, wherein the labeling molecule has the general structure :

T-ZD-A-

wherein ZD is the zwitterionic dye moiety, T is the titratable moiety, and A is linker moiety.

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12. The labeling molecule of Claim 1, wherein the labeling molecule has the general structure :

ZD-T-A-

wherein ZD is the zwitterionic dye moiety, T is the titratable moiety, and A is linker moiety.

13. The labeling molecule of Claim 2, wherein the labeling molecule has the general structure :

T-ZD-C-A-

wherein ZD is the zwitterionic dye moiety, T is the titratable moiety, C is the cleavable moiety,
5 and A is linker moiety.

14. The labeling molecule of Claim 3, wherein the labeling molecule has the general structure :

T-ZD-C-I-A-

wherein ZD is the zwitterionic dye moiety, T is the titratable moiety, C is the cleavable moiety, I is
10 the stable isotope moiety and A is linker moiety.

15. The labeling molecule of Claim 3, wherein the labeling molecule has the general structure :

ZD-T-C-I-A-

wherein ZD is the zwitterionic dye moiety, T is the titratable moiety, C is the cleavable moiety, I is
the stable isotope moiety and A is linker moiety.

15 16. A target protein labeled with the labeling molecule of Claim 1, wherein the linker is covalently
attached to the target protein.

17. A method of labeling a target protein comprising the steps of:

a. providing an optical labeling molecule comprising

- i. a zwitterionic dye moiety;
- ii. a titratable group moiety;
- iii. an optional cleavable moiety; and
- iv. a functional linker moiety;

b. contacting the target protein with the labeling molecule to form a labeled protein.

25 18. A method according to Claim 17 wherein a plurality of target proteins are each labeled with a
different labeling molecule.

19. A method of performing protein analysis on a plurality of proteins comprising:

a. providing a plurality of different labeled proteins, each comprising a

30 i. a different zwitterionic dye moiety;

- ii. a titratable group moiety; and
 - iii. an optional cleavable moiety;
 - b. determining the presence or absence of each of the different labeled proteins.
- 5 20. A method according to Claim 19 wherein the plurality of different labeled proteins are mixed and separated simultaneously prior to the determining the presence or absence of each of the different labeled proteins in the samples.
- 10 21. A method according to Claim 20 wherein the different labeled proteins are separated by a method selected from the group consisting of 1D gel electrophoresis, 2D gel electrophoresis, capillary electrophoresis, 1D chromatography, 2D chromatography, 3D chromatography, and mass spectroscopy.
- 15 22. A method according to Claim 19 further comprising the step of determining the relative quantity of the different labeled proteins.
- 20 23. A method according to Claim 19 wherein the cleavable moiety is present, the method further comprising cleaving the cleavable moiety to remove the labeling molecule from the different labeled proteins.
- 25 24. A method according to Claim 23 wherein the identities of the separated proteins are determined by mass spectral techniques after removal of the dye tags.
- 30 25. A method according to Claim 19 wherein the cleavable moiety is present, each of the labeled proteins further comprising a different stable isotope tag moiety located between the functional linker moiety and the cleavable moiety.
- 35 26. A method according to Claim 25 further comprising the steps of cleaving the cleavable moiety to produce isotope labeled proteins and determining the quantity of the isotope labeled proteins.
- 40 27. A method according to Claim 26 wherein the identity of the isotope labeled proteins is determined by mass spectral techniques.